American Journal of Emergency Medicine 34 (2016) 370–374

Contents lists available at ScienceDirect

American Journal of Emergency Medicine

journal homepage: www.elsevier.com/locate/ajem

Original Contribution

Inflammatory biomarkers predicting prognosis in patients with acute dyspnea☆,☆☆,★

Karolin Wiklund, MD [a,*], Klas Gränsbo, MD [b], Nathalie Lund, MD [b], Marjaneh Peyman [b], Lena Tegner [b], Maria Toni-Bengtsson [b], Mattias Wieloch, MD, PhD [a], Olle Melander, MD, PhD [a]

[a] The Department of Clinical Sciences Malmo, Faculty of Medicine, Lund University, Lund, Sweden
[b] Department of Internal Medicine and Emergency Medicine, Skane University Hospital, Malmo, Sweden

ARTICLE INFO

Article history:
Received 6 September 2015
Received in revised form 23 October 2015
Accepted 30 October 2015

ABSTRACT

Objective/Purpose: The objective was to identify inflammatory biomarkers that predict risk of 90-day mortality in patients with acute dyspnea.
Method: We analyzed 25 inflammatory biomarkers, in plasma, in 407 adult patients admitted to the emergency department (ED) with acute dyspnea and related them to risk of 90-day mortality using Cox proportional hazard models adjusted for age, sex, oxygen saturation, respiratory rate, C-reactive protein, and Medical Emergency Triage and Treatment System–Adult score.
Results: Fifty patients (12%) died within 90 day from admission. Two strong and independent biomarker signals were detected: The hazard ratio (95% confidence interval) for 90-day mortality per 1-SD increment of interleukin-8 (IL-8) was 2.20 (1.67-2.90) ($P = 2.5 \times 10^{-8}$) and for growth differentiation factor–15 (GDF-15) was 3.45 (2.18-5.45) ($P = 1.3 \times 10^{-7}$) A Biomarker Mortality Risk Score (BMRS) summing standardized and weighted values of IL-8 and GDF-15 revealed that of patients belonging to quartile 1 (Q1) of the BMRS, only 1 patient died, whereas 32 patients died among those belonging to quartile 4. Each 1-SD increment of the BMRS was associated with a hazard ratio of 3.79 (2.50-5.73) ($P = 2 \times 10^{-10}$) for 90-day mortality, and the point estimate was 13 times higher in Q4 as compared with Q1 of the BMRS (P_{trend} over quartiles $= 2 \times 10^{-6}$).
Conclusion: Interleukin-8 and GDF-15 are strongly and independently related to risk of 90-day mortality in unselected patients admitted to the ED because of acute dyspnea, suggesting that they may guide first-line physicians at the ED in risk assessment which in turn could lead to more accurate level of care and treatment intensity.

1. Introduction

1.1. Background

Dyspnea is one of the most common symptoms at the emergency department (ED), accounting for approximately 3-4 million visits in the ED every year in the United States [1]. The underlying differential diagnoses are diverse and range from conditions with high risk of mortality to virtually no risk of mortality at all. Chronic diseases such as congestive heart failure (CHF) and chronic obstructive pulmonary disease (COPD) are common in patients with acute dyspnea as well as infections, pulmonary embolism, and anxiety disorders. Physical examination, history, laboratory work, and imaging techniques are used in assessing the cause [2]. However, the identification of the correct diagnosis is just one step toward correctly treating the patient, and sometimes, the main underlying cause of acute dyspnea is not unmasked until during hospitalization or after the patient has left the ED. An equally important task for the physician at the ED is to make decisions of intensity of treatment and level of care, or follow-up in the case of hospital discharge. One of the key factors to take into account as a basis for these decisions is the patient's risk of long-term mortality.

Not only in infections but also in chronic diseases, for example, CHF and COPD, has inflammation been suggested be a part of the pathological processes [3,4]. By definition, inflammation is a protective tissue response to injury or destruction of tissues, which aims at eliminating the injurious agent and the injured tissues. Clinically and epidemiologically, inflammation is usually estimated by measurement of circulating substances that are released as a cause or consequence of an inflammatory response, and the most widely used biomarker for inflammation is C-reactive protein (CRP). However, inflammation is a complex process,

☆ Funding: This work was supported by grants from the European Research Council (StG-282255), the Swedish Medical Research Council, the Swedish Heart and Lung Foundation, the Medical Faculty of Lund University, Malmö University Hospital, the Albert Påhlsson Research Foundation, the Crafoord Foundation, the Ernhold Lundströms Research Foundation, Region Skane, the Hulda and Conrad Mossfelt Foundation, the King Gustaf V and Queen Victoria Foundation, the Novo Nordisk Foundation, and the Wallenberg Foundation.
☆☆ Conflicts of interests: none.
★ Presented as poster presentation at European Society of Cardiology Congress on Heart Failure, Seville, Spain, on May 24, 2015.
* Corresponding author at: CRC Hus 92 plan 11, Jan Waldenströms gata 35, 21428, Malmo, Sweden.
E-mail address: karolin.wiklund@med.lu.se (K. Wiklund).

http://dx.doi.org/10.1016/j.ajem.2015.10.052

and the total variance of the wide range of inflammatory substances can only partially be explained by CRP.

Although the main cause of dyspnea is usually established within a few days of hospitalization, the physician in the ED is commonly subjected to a patient with little information except brief medical history and a state of acute dyspnea. Even in cases when the patient has one or several known diseases which can cause acute dyspnea, for example, CHF, COPD, and previous pneumonia, the main underlying cause and severity of the current episode could be difficult to assess. Therefore, we chose to study an unselected patient population with acute dyspnea.

The aim of our study was to investigate if inflammatory biomarkers predict 90-day mortality in this patient group.

The choice of inflammatory biomarkers was based on the hypothesis that inflammation may have a central role in many of the more malignant underlying causes to acute dyspnea and that inflammatory biomarkers therefore may be particularly useful to predict long-term prognosis in this patient category.

2. Method

2.1. Study population

The study was conducted at the ED at the Skåne University Hospital in Malmö (SUS Malmö), which serves a catchment area of approximately 400,000 inhabitants and has almost 85,000 annual visits. Adult patients that presented to the ED with the main symptom of acute dyspnea between the 6th of March 2013 and the 29th of May 2014 were offered to participate in the study. Patients were only included during office hours, 6:45 AM to 4:30 PM, when a research nurse was present. All patients were triaged to Medical Emergency Triage and Treatment System–Adult score (METTS-A) category 1-4 (see explanation of METTS-A below) and had routine blood chemistry performed.

Four-hundred thirty-nine patients were enrolled in the study. Of these, 32 were excluded because of missing values of one of more of the covariates used in the statistical analyses (see below), yielding a total number of 407 patients that were included in the current study.

Medical Emergency Triage and Treatment System–Adult was measured according to standard procedures which was the standard tool for triaging patients during the time of inclusion [5]. Medical Emergency Triage and Treatment System–Adult is a 5-level triage tool combining vital signs (oxygen saturation, respiratory rate, heart rate, blood pressure, degree of consciousness, and body temperature) and symptoms. The 5 levels are as follows: red (priority 1), life threatening; orange (priority 2), potentially life threatening; yellow (priority 3), not life threatening but in need of medical attention within 2 hours; green (priority 4), not life threatening and not in need of immediate care; and blue (priority 0), not in need of emergency care and should be referred to primary care.

2.2. Clinical parameters

Respiratory rate was manually counted. Blood pressure, oxygen saturation, and heart rate were measured with a fully automated oscillometric device (CARESCAPE Monitor B850 or B650, General Electric Healthcare) [6,7], and degree of consciousness was determined according to the Reaction Level Scale +[8]. After inclusion, patients were asked about smoking habits in 3 categories (never smoked, former smoker, and active smoker which included occasional smoker), prevalence of chronic diseases known to cause dyspnea (eg, CHF, COPD, asthma, coronary artery disease, atrial fibrillation, restrictive lung disease, cancer, thromboembolic disease, rheumatic disease), and current medication. Patient journals were examined by the research nurses to confirm previous diagnoses. Plasma concentration of CRP was measured using a Radiometer ABL800 Flex machine [9] or Afinion AS100 Analyzer System [10]. Blood samples were collected immediately at presentation with separation of serum and plasma and were frozen and stored at −80°C for later analysis of biomarkers.

2.3. Biomarker selection and measurement

The frozen blood samples were analyzed by the Proseek Multiplex CVD I biomarker panel (Olink Bioscience, Uppsala, Sweden) which includes the 29 selected inflammatory markers chosen for revision in the current study (Table 2). The method is a multiplex immunoassay based on a Proximity Extension Assay [11]. For details, please see the supplementary appendix.

Of the 29 selected inflammatory biomarkers, 4 were excluded because of chemically undetectable levels of the specific biomarker on >98% of the collected samples.

All assay characteristics including detection limits and measurements of assay performance and validations are available from the manufacturer's Web page [12].

The end point was all-cause mortality within 90 days from presentation to the ED. The personal identification number was linked to the national civil registry, in which survival status up to 90 days after presentation was retrieved. If the patient had died, the date of death was recorded.

2.4. Statistical analysis

Because of skewed distributions of the biomarkers, the natural logarithms were derived for the biomarker's values and the values of CRP to achieve statistical normality, and the log-transformed values were expressed on a standardized scale (per 1-SD increment). Cox proportional hazard models were created to relate each biomarker to the 90-day mortality using time of presentation as the start of the follow-up time and time of death as the end of follow-up if death occurred or to the end of the study (90 days after presentation) if death did not occur. Model 1 was adjusted for sex and age. The biomarkers with a P value < .05 were then individually entered in model 2 which was adjusted for sex, age, METTS-A, oxygen saturation, respiratory rate, and CRP. Biomarkers that had P value < .05 in model 2 were entered simultaneously in the Cox proportional hazard model and adjusted for the same factors as in model 2. Biomarkers which were significant in this analysis and were also retained at a P value of < .05 in both forward and backward selection were summed up to a score and weighted using their β-coefficients from the Cox proportional hazard model (ie, β-coefficient for biomarker 1 × z score of biomarker 1 + β-coefficient of biomarker 2 × z score of biomarker 2, etc). The sum of these weighted values of biomarkers was in turn expressed as multiples of 1 SD (z score) and was ranked and ordered into quartiles. Additional adjustments on top of model 2 were performed for comorbidities, smoking (current, former, and never smoker) and the biomarker N-terminal of the prohormone brain natriuretic peptide (NTproBNP). A P value of < .05 was considered statistically significant. The data were statistically analyzed using IBM SPSS statistics 22 (SPSS Inc, Chicago, IL).

The study was approved by the regional ethics board of Lund, Sweden, and followed the precepts established by the Declaration of Helsinki.

Table 1
Clinical characteristics of study population

Age (y) mean ± SD	70.0 ± 18.3
Sex, female (%)	213 (52.3)
METTS-A[a] (%)	46 (11.3)/137 (32.7)/186 (45.7)/38 (9.3)
Oxygen saturation (%) mean ± SD	93.8 ± 6.1
Respiratory rate mean ± SD	23.7 ± 6.8
CRP (mg/dL) median (IQR)	8.4 (3.4-34.0)
CHF n (%)	145 (35.6)
COPD n (%)	112 (27.5)
Smoking[b] n (%)	107 (25.3)/214 (52.6)/77 (18.9)

[a] Four categories of METTS-A priority: 4/2/3/1, least critical to most critical.
[b] Three categories: nonsmokers/former smokers/active smokers.

3. Results

Clinical characteristics of the study population are shown in Table 1. The mean age of the study population was 70.0 ± 18.3 years, 52.3% were women, and the mean respiratory rate was 24 ± 7 min^{-1}. Two hundred forty-seven patients were admitted to a ward; of these, 88 were admitted to a monitored ward (intensive care unit, coronary care unit, or acute medical ward). During follow-up, 50 patients died; 44 of these had been admitted to a ward at the time of presentation. In Table 2, the result of model 1 (adjusted for sex and age) and model 2 (adjusted for sex, age, METTS-A, oxygen saturation, respiratory rate, and CRP) is presented. Twenty of the biomarkers showed a significant association with 90-day mortality and were subsequently entered in model 2. In model 2, 16 of the biomarkers were significantly associated with risk of 90-day mortality. Using model 2 adjustments, the 16 biomarkers were simultaneously entered into the Cox proportional hazard models by forcing all 16 markers into the model as well as using both forward and backward stepwise selection. Two biomarkers, interleukin-8 (IL-8) and growth differentiation factor–15 (GDF-15), showed significant and independent association with 90-day mortality in all 3 analyses. Each 1-SD increment of IL-8 was associated with increased risk of 90-day mortality with a hazard ratio (HR) (95% confidence interval [CI]) of 2.20 (1.67-2.90) ($P = 2.5 \times 10^{-8}$), and GDF-15 conferred an HR (95% CI) of 3.45 (2.18-5.45) ($P = 1.3 \times 10^{-7}$). Apart from these 2 biomarkers, age per year (1.04 [1.01-1.07], $P = 0.007$) and respiratory rate (per min^{-1}) (1.08 [1.03-1.13], $P = 0.002$) significantly predicted 90-day mortality. C-reactive protein was not significantly associated with risk of 90-day mortality (HR [95% CI], 0.99 [0.72-1.35]; $P = 0.94$).

The summed biomarker mortality risk score of IL-8 and GDF-15 (BMRS) was strongly associated with risk of 90-day mortality, with each 1-SD increment of the BMRS conferring an almost 4-fold increase of the relative risk for death (Table 3). As shown in Table 3 and the Figure, quartile analyses revealed that mortality was very low in subjects with values less than the median of the summed biomarker score and increased steeply over quartiles 3-4. In quartile 1, the 90-day mortality was only 1%, whereas it increased to 32% in quartile 4.

Smoking, history of CHF, history of COPD, and the biomarker NT-proBNP were added to model 2 adjustments, but this did not affect the relationship between the BMRS and 90-day mortality. N-terminal of the prohormone brain natriuretic peptide was not significantly associated with 90-day mortality in this study population.

4. Discussion

The key findings of the current study was that IL-8 and GDF-15, measured in plasma collected immediately upon entry to the ED, individually and in aggregate strongly and independently predict risk of 90-day mortality in patients with acute dyspnea. In patients with values less than the median values of the BMRS (ie, sum of standardized values of IL-8 and GDF-15), mortality was only 2%, whereas it was 32% in patients in the top quartile, with a multivariate adjusted relative risk of 13 in top vs bottom quartile of the BMRS. This demonstrates for the first time that inflammatory biomarkers can predict mortality in patients presenting with acute dyspnea irrespective of underlying pathology. Whereas there are previous studies examining the prognostic value of few selected biomarkers in CHF [13] and lower respiratory tract infections [14], we are not aware of any prior study on a broad set of inflammatory biomarkers, taken upon presentation to the ED, in unselected patients with acute dyspnea. It is also important to recognize that the BMRS and 90-day mortality were not affected by adjustment for prevalence of COPD and CHF in this study, making the BMRS more important than current pathology in assessing prognosis.

Since the *P* values for the BMRS where shown to be solid and withstand using of the Bonferroni correction, we do not think multiple comparisons when testing 25 different inflammatory biomarkers have flawed our key results. The weakest *P* value (*P* for trend over quartiles of the BMRS) gives us a corrected $P_{\text{Bonnferroni}} = 2 \times 10^{-6} \times 25 = 5 \times 10^{-5}$, demonstrating that the significance of the results is solid.

Along with an aging population, which inevitably leads to increased occurrence of both acute and chronic diseases, there is an escalating demand on the health care system to use its resources more efficiently without compromising medical safety and quality of care. As the main

Table 2
Relationships between inflammatory biomarkers and risk of 90-day mortality

Biomarker	HR (95% CI); age and sex adjusted	*P* value	HR (95% CI); age, sex, METTS-A, oxygen saturation, respiratory rate, and CRP adjusted	*P* value
CCL4	1.41 (1.14-1.76)	1.9×10^{-3}	1.34 (1.08-1.66)	8.0×10^{-3}
CCL20	1.80 (1.36-2.40)	4.2×10^{-5}	1.52 (1.10-2.11)	1.2×10^{-2}
CD40-L	1.22 (0.91-1.64)	NS		
CHI3L1	1.70 (1.16-2.51)	7.0×10^{-3}	1.45 (0.95-2.22)	NS
CXCL1	1.57 (1.20-2.07)	1.2×10^{-3}	1.47 (1.10-1.94)	8.1×10^{-3}
ECP	1.28 (0.93-1.77)	NS		
SELE	1.25 (0.94-1.70)	NS		
CX3CL1	1.38 (1.01-1.87)	4.2×10^{-2}	1.38 (1.03-1.84)	3.2×10^{-2}
FS	1.98 (1.41-2.78)	7.5×10^{-5}	1.83 (1.28-2.61)	8.9×10^{-4}
GAL3	1.63 (1.25-2.13)	3.4×10^{-4}	1.59 (1.21-2.08)	7.5×10^{-4}
GDF15	3.51 (2.29-5.38)	7.9×10^{-9}	3.45 (2.18-5.45)	1.3×10^{-7}
hK11	1.47 (1.10-1.95)	8.5×10^{-3}	1.54 (1.16-2.03)	2.7×10^{-3}
IL-1-RA	1.49 (1.11-2.00)	8.1×10^{-3}	1.19(0.83-1.72)	NS
IL-6	1.99 (1.44-2.76)	3.7×10^{-5}	1.72(1.16-2.57)	7.1×10^{-3}
IL-6-RA	1.47 (1.07-2.02)	1.8×10^{-2}	1.50 (1.08-2.08)	1.5×10^{-2}
IL-8	2.29 (1.80-2.91)	1.1×10^{-11}	2.20 (1.67-2.90)	2.5×10^{-8}
IL-16	1.28 (0.95-1.73)	NS		
IL-18	1.42 (1.10-1.82)	6.6×10^{-3}	1.27 (0.97-1.65)	NS
IL-27-A	1.97 (1.38-2.81)	2.0×10^{-4}	1.69 (1.13-2.53)	1.0×10^{-2}
LOX-1	1.90 (1.36-2.65)	1.8×10^{-4}	1.68 (1.18-2.40)	4.4×10^{-3}
MCP1	1.60 (1.21-2.12)	1.0×10^{-3}	1.52(1.16-2.01)	2.8×10^{-3}
NEMO	1.34 (1.01-1.78)	4.6×10^{-2}	1.21 (0.90-1.63)	NS
RAGE	1.16 (0.85-1.59)	NS		
ST2	3.42 (2.22-5.27)	2.6×10^{-8}	3.22 (1.96-5.29)	4.2×10^{-6}
U-PAR	2.46 (1.64-3.69)	1.3×10^{-5}	2.12 (1.37-3.25)	7.1×10^{-4}

C-C motif chemokine 4 (CCL4), C-C motif chemokine 20 (CCL20), CD40-ligand (CD40-L), chitinase-3-like protein 1 (CHI3L1), C-X-C motif chemokine 1 (CXCL1), eosinophil cationic protein (ECP), E-selectin (SELE), fractalkine (CX3CL1), follistatin (FS), galectin-3 (GAL3), growth differentiation factor 15 (GDF15), human kallikrein 11 (hK11), interleukin 1 receptor antagonist protein (IL-1-RA), interleukin 6 (IL-6), interleukin 6 receptor subunit alpha (IL-6-RA), interleukin 8 (IL-8), interleukin 16 (IL-16), interleukin 18 (IL-18), interleukin 27 subunit alpha (IL-27-A), lectin-like oxidized LDL receptor 1 (LOX-1), monocyte chemotactic protein 1 (MCP1), NF-kappa-B essential modulator (NEMO), receptor for advanced glycosylation end products (RAGE), ST2 protein (ST2), urokinase plasminogen activator surface receptor (U-PAR).

Table 3
Multivariate adjusted HRs for the biomarker mortality score in relation to 90-day mortality

	All patients	*P* value	Quartile 1	Quartile 2	Quartile 3	Quartile 4	*P* trend
N/n events	407/50		101/1	102/2	102/14	102/32	
HR (95% CI)[a]	3.79 (2.50-5.73)	3.2×10^{-10}	1 (Ref)	0.91 (0.08-10.56)	5.25 (0.63-43.62)	13.17 (1.60-108.46)	2.0×10^{-6}

[a] Adjusted for age, sex, METTS-A, oxygen saturation, respiratory rate, and CRP.

hospital inflow of acutely ill patients takes place at the ED, risk stratification tools which may help the first-line physician at the ED to decide on level of care, as well as the correct level of follow-up, are becoming increasingly important. In this context, biomarkers are attractive candidates given their relatively low cost, their potential for generating results fast, and that patients are not exposed to radiation or other risks.

Our findings have several potential clinical implications. The mortality within 90 days in the patient population was high (12%). Given the relatively short follow-up time, deaths occurring within that time frame have a high likelihood of having a relationship with the acute illness underlying the acute dyspnea for which the patients sought help. Thus, risk of long-term mortality is likely to be a good proxy of the severity of the acute underlying illness. Given the large difference in 90-day mortality between patients in quartiles 1-2 and quartile 4 of the BMRS (Figure), measurement of IL-8 and GDF-15, upon presentation to the ED, in patients with acute dyspnea may be a valuable complementary support for the first-line physician to make decisions to safely send home patients with low values of the BMRS and provide follow-up in primary care or no follow-up at all. Furthermore, as one-third of patients in the top quartile of the BMRS died within 90 days, such high values may support prioritization of more intensive care. In addition, given the high 90-day mortality in patients with high BMRS values, such patients may require extra attention after being discharged from the hospital or, in the presence of no clinical indication for immediate admission, a close follow-up at a hospital outpatient clinic. Importantly, the BMRS performed much better than and was statistically independent of initial vital signs, CRP, comorbidities, and the triage algorithm score used (METTS-A), the latter of which has previously been shown to predict in-hospital mortality [5].

We focused the study on unselected patients with acute dyspnea rather than at selected groups of patients with a specific main diagnosis (eg, CHF, COPD, or infection). The reason for doing this was that we wanted to identify biomarkers which are helpful upon immediate presentation at the ED. This is when the first-line physician who meets the patient commonly has limited or no access at all to what may be the underlying main diagnosis.

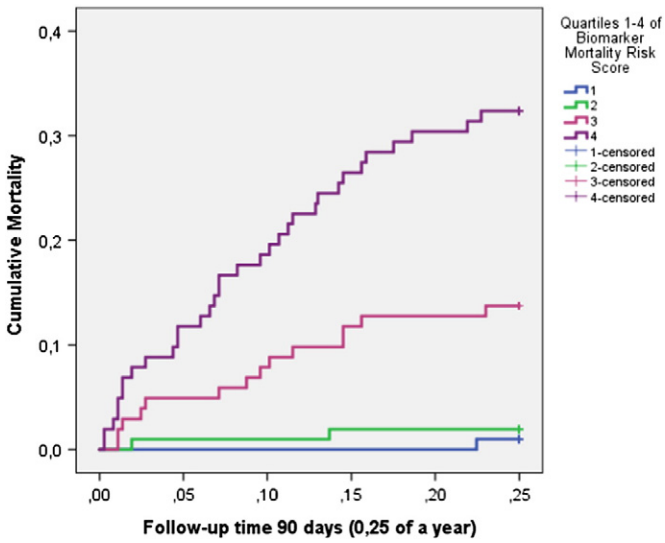

Figure. Kaplan-Meier plot showing cumulative mortality during 90 days of follow-up. Quartile 1 denotes the lowest values of the BMRS; and Quartile 4, the highest values.

All of the conditions underlying acute dyspnea have different treatment plans, making it important to find the underlying cause. The BMRS may motivate further examination and extensive medical testing. For example, in a patient with a high BMRS, it may be more important to use more advanced examinations early to find the exact underlying cause, possibly staging and beginning short-term and long-term treatment quickly. In a patient with a low BMRS, such urgency is lower, and further examinations could be decided upon later on and when needed in primary care. This would aid in prioritizing and distributing the time assets and monetary assets of the ED, and also in health care in general, and studies specifically addressing these aspects are warranted in the future.

Interleukin-8, also known as *chemokine (C-X-C motif) ligand 8*, is produced by macrophages, monocytes, fibroblasts, epithelial cells, and endothelial cells. It induces chemotaxis for naive T cells and mobilizes and activates neutrophils and angiogenesis [15]. It is produced quickly as a reaction and progressor of inflammation. It is already in some clinical use as an early marker of neonatal sepsis and systemic inflammation [16].

Growth differentiation factor–15 is part of the transforming growth factor β family (TGF-β superfamily). It is known under many names, for example, macrophage inhibitory cytokine, NSAID-activated gene, prostate-derived factor, placental bone morphogenetic protein, placental TGF-β, and PL74. It is a regulatory protein in the inflammatory pathway. Growth differentiation factor–15 is found in all tissues but is only physiologically active in the placenta where it is thought to ensure the survival of the fetus by inhibiting the mother's immune response. It is however present and active in many pathologies, for example, cancer, cardiopulmonary disease, renal failure, diabetes, and congenital anemia. A high level of GDF-15 has been associated with an increased risk of cardiovascular events and with more malignant phenotypes of many tumors [17]. Growth differentiation factor–15 has been shown to be associated with a higher mortality irrespective of the cause of death [18]. It is not yet in clinical use.

According to the latter facts on the biomarkers in addition to our data, inflammation is a contributor or indicator of the mortality in the study population. The inflammation in our study population could however not be shown by using the traditional CRP. C-reactive protein is increased in systemic inflammation, viral infections, and bacterial infections and is particularly high in sepsis [15]. Our results do not preclude CRP from being important in aiding in the diagnostic process; however, its prognostic value in terms of 90-day mortality in acute dyspnea patients seems limited.

4.1. Limitations

Both COPD and CHF are characterized by increased inflammatory activity, and they are both associated with markedly reduced life expectancy and commonly present themselves with dyspnea. As stated previously, we were therefore surprised that we could show no relationship between 90-day mortality and the presence of CHF or COPD. However, we acknowledge that the information available in medical records combined with the patient's own reported medical history may not be fully accurate and does not take into account factors of importance for survival such as duration and severity of these diseases [19,20]. This is, however, a limitation which is also commonly present

in the real medical setting at the ED, that is, the setting in which we propose the biomarkers to be used.

In the current study, we cannot draw any conclusions as to whether IL-8 and GDF-15 are bystanders, correlating with the underlying pathophysiology or whether they may be causally involved in the acute disease process.

Although the nominal differences of numbers of deaths and point estimates of HRs in top vs bottom quartiles were large, one must remember that the CIs were wide, and thus the point estimates should be interpreted with caution. This is a consequence of relatively low numbers of patients and events in each quartile. On the other hand, in the analyses of the continuous relationship between the BMRS (as well as IL-8 and GDF-15 individually) and the risk of 90-day mortality, where the average risk per 1-SD increase was calculated, the statistical significance was strong and CIs narrower. Hence, we suggest that the BMRS does provide strong independent information on 90-day mortality risk. However, larger multicenter studies are warranted to define the most clinically important cutoff values (top and bottom quartiles or quintiles etc)

Ninety days is a commonly used follow-up time in ED-based biomarker studies; it may intuitively be regarded as a long time to evaluate mortality in acute conditions. In-hospital mortality is one alternative end point, which however does not standardize the follow-up time, and 30-day mortality is another alternative end point, both of which are likely to have a relation to the acute illness underlying the dyspnea. On the other hand, if we had chosen a shorter follow-up time, that would have been at the expense of missing some deaths which indeed were related to the acute illness presenting as dyspnea at the ED. The Kaplan-Meier plot (Figure) shows an almost constant proportionality over 90 days, suggesting that the HR would have been equally high using a 30-day end point.

A relatively long follow-up time would be particularly important when considering the potential clinical implication of using the BMRS to identify patients with low values and low risk and using this as support to safely discharge them with little or no need for follow-up. Ninety days of follow-up therefore appears to be a decent compromise both to guide decisions prioritizing higher level of care or need of close short-term follow-up and to support safe discharge.

5. Conclusion

Our results show that a biomarker risk score of IL-8 and GDF-15 is a strong predictor of 90-day mortality in patients in an emergency setting with acute dyspnea, independently of vital signs, triage priority, CRP, and important comorbidities. Therefore, Biomarker Risk Score (BMRS) is suggested to be used at the ED to identify patients with a higher need of medical attention and management. It may also turn out to be useful to support the ED physician's decision to safely discharge patients with a low risk of mortality.

References

[1] Parshall MB, Schwartzstein RM, Adams L, Banzett RB, Manning HL, Bourbeau J, et al. An official American Thoracic Society statement: update on the mechanisms, assessment, and management of dyspnea. Am J Respir Crit Care Med 2012;185:435–52.
[2] Ferrin MS, Tino G. Acute dyspnea. AACN Clin Issues 1997;8:398–410.
[3] Sharma K, Kass DA. Heart failure with preserved ejection fraction: mechanisms, clinical features, and therapies. Circ Res 2014;115:79–96.
[4] Angelis N, Porpodis K, Zarogoulidis P, Spyratos D, Kioumis I, Papaiwannou A, et al. Airway inflammation in chronic obstructive pulmonary disease. J Thorac Dis 2014; 6(Suppl. 1):S167–72.
[5] Widgren BR, Jourak M. Medical Emergency Triage and Treatment System (METTS): a new protocol in primary triage and secondary priority decision in emergency medicine. J Emerg Med 2011;40:623–8.
[6] Carescape Monitor B650. Accessed 11/11, 2014, at http://www3.gehealthcare.com/en/products/categories/patient_monitoring/patient_monitors/carescape_monitor_b650#tabs/tab79DF9315AB9B4FDAB9F99A95D3C2677D; 2014.
[7] Carescape Monitor B850. Accessed 11/11, 2014, at http://www3.gehealthcare.com/en/products/categories/patient_monitoring/patient_monitors/carescape_monitor_b850; 2014.
[8] Starmark JE, Stalhammar D, Holmgren E. The Reaction Level Scale (RLS85). Manual and guidelines. Acta Neurochir 1988;91:12–20.
[9] ABL800 FLEX analyzer. Accessed 11/11, 2014, at http://www.radiometeramerica.com/~/media/Files/RadiometerComCloneset/RAME/Brochures/Products/ABL800%20specs.pdf; 2011.
[10] Afinion AS100 Analyzer System. Accessed 11/11, 2014, at http://www.afinion.net/specification; 2009.
[11] Lundberg M, Eriksson A, Tran B, Assarsson E, Fredriksson S. Homogeneous antibody-based proximity extension assays provide sensitive and specific detection of low-abundant proteins in human blood. Nucleic Acids Res 2011;39, e102.
[12] Proseek Multiplex CVD I 96x96. at http://www.olink.com/sites/default/files/0969%20v1.1%20Proseek%20Multiplex%20CVD%20I%20Data%20Package_final.pdf; 2013.
[13] Gori CS, Magrini L, Travaglino F, Di Somma S. Role of biomarkers in patients with dyspnea. Eur Rev Med Pharmacol Sci 2011;15:229–40.
[14] Schuetz P, Christ-Crain M, Thomann R, Falconnier C, Wolbers M, Widmer I, et al. Effect of procalcitonin-based guidelines vs standard guidelines on antibiotic use in lower respiratory tract infections: the ProHOSP randomized controlled trial. JAMA 2009;302:1059–66.
[15] Murphy K, Travers P, Walport M, Janeway C. Janeway's immunobiology. 8th ed. New York: Garland Science; 2012.
[16] Truedsson L. Klinisk immunologi. Lund: Studentlitteratur; 2012.
[17] Corre J, Hebraud B, Bourin P. Concise review: growth differentiation factor 15 in pathology: a clinical role? Stem Cells Transl Med 2013;2:946–52.
[18] Wiklund FE, Bennet AM, Magnusson PK, Eriksson UK, Lindmark F, Wu L, et al. Macrophage inhibitory cytokine–1 (MIC-1/GDF15): a new marker of all-cause mortality. Aging Cell 2010;9:1057–64.
[19] Shavelle RM, Paculdo DR, Kush SJ, Mannino DM, Strauss DJ. Life expectancy and years of life lost in chronic obstructive pulmonary disease: findings from the NHANES III Follow-up Study. Int J Chron Obstruct Pulmon Dis 2009;4:137–48.
[20] Ahmed A, Aronow WS, Fleg JL. Higher New York Heart Association classes and increased mortality and hospitalization in patients with heart failure and preserved left ventricular function. Am Heart J 2006;151:444–50.